

origin of pigment cells in amphibians (Dushane, 1935), birds (Dorris, 1938) and mammals (Rawles, 1947). It merely signifies that under certain abnormal conditions, other than the usual cells can take over the function of pigment cell formation.

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### Regeneration of Neural Retina and Lens from Pigment Cells in the Eyes of Adult Salamanders.\*

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The neural retina of adult salamander eyes degenerates when deprived of its blood supply either by severing the vessels which vascularize it (Stone and Chace, 1941) or by transplanting the entire eye (Stone and Zaur, 1940; Stone and Cole, 1943; Stone and Ellison, 1945). The only retinal cells which survive are those of the pigmented epithelial layer from which a new neural retina regenerates (Stone, 1949). This has been studied in detail in many grafted adult urodele eyes. Degeneration spreads throughout the neural retina during the first three weeks. It often proceeds at different rates in various parts of the same eye, allowing early stages of regeneration to begin in some areas

while adjacent regions are still degenerating. The true origin of the new neural retina is therefore more easily followed in cases where degeneration is simultaneous in all parts or where the entire neural retina is detached from the underlying retinal pigment layer by a gentle stream of Ringer's solution and then removed intact through a broad dorsal slit in the eye.

Degeneration of the neural retina in grafted eyes usually spreads rapidly until the retinal pigment cells are finally denuded and the cellular debris is absorbed above them. The pigment cells then become dense black oval or flattened bodies resembling those found soon after surgical removal of the entire neural retina. Now they take on temporarily another function, entering the critical stages which can be followed step by step as they determine the origin of the new neural retina.

These pigment cells increase markedly in size. Their nuclei become distinct as they undergo mitosis. One daughter cell migrates inward, loses its pigment and with similar ones forms a sharply defined layer, which by further cell division gives rise to a new neural retina. The other daughter cell retains its pigment and later takes on the status of a retinal pigment cell when the neural retina above it has fully regenerated. The functional capacity of this cell is finally expressed by the migration of its pigment granules in a light-adapted eye as soon as the rods and cones above it are differentiated.

When a small portion of the neural retina is excised, or is detached as a permanent elevated fold, regeneration is also called forth from the underlying retinal pigment cells. On the other hand the initial reaction of the pigment cell to a small retinal injury is one of rapid mitosis and mass migration into the wound, somewhat similar to that found in the retinal wounds of other vertebrates. However, in the urodele retina these cells later lose their pigment and differentiate into neural retinal tissue. Retinal pigment epithelium transplanted into the eye chambers also gives rise to neural retina.

Since the early pigment cell changes involve replacement of a lost tissue the term "dedifferentiation" might be considered as it applies to them. This term is often loosely and obscurely applied to cells which are assumed to be taking part in regeneration, although up to that moment they are recognized as highly differentiated elements with special morphological and functional characteristics. Under the proper stimulus they are supposed to lose their special features and take on a role of supplying new cells that will later develop into similar or different elements. If the term is defined in this sense it can be said that the retinal pigment cell in the salamander eye is capable of dedifferentiation.

Another pigment cell in the eye of *Triturus* salamanders may be considered in the same category, for when the lens is removed the

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heavily pigmented cells along the free pupillary margin of the dorsal iris increase in size, become depigmented, undergo mitosis and form a vesicle that develops into a lens (Wachs, 1914, and others). When the latter becomes detached the cells in the dorsal iris regain their normal appearance. If varying amounts of dorsal iris are replaced by non-lens regenerating ventral iris, two widely separated lenses develop from the remaining dorsal pupillary margins after removal of the original lens. Secondary pupils experimentally produced in various regions of the dorsal iris by the insertion of pieces of pliciform or cornea show that potentiality for lens formation is quite widely distributed and not confined to the free pupillary margin (Stone and Vultee, 1949).

These changes in the pigment cells can be experimentally inhibited by the following: 1) the presence of a transplanted normal lens of the same or another species (Stone, 1945); 2) the presence of a thirty-day lens regenerate (Stone, 1943); 3) injections of aqueous humor from eyes containing lenses (Stone and Vultee, 1949); 4) the presence of some carcinogens.

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#### X-Ray Effects on Mouse Pigmentation as Related to Melanoblast Distribution.

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Several features of the x-ray-induced greying response suggest certain properties of the pigmentation system. Follicles react largely as independent units, producing fully pigmented hairs (no apparent gross effect), white hairs, or infrequently mosaics. The effect is permanent or toward more white hairs in successive hair generations of the particular follicle (Chase, Quastler, and Skaggs, 1947; Chase, 1949). The percentage of follicles producing only white hairs after treatment increases with dose (200-1000 r) and with dose-rate. The percentage for a given dose is greater for follicles treated in catagen and telogen stages and for follicles of the smaller hair types (Chase, 1949; Chase and Rauch, in press). In a mosaic some of the medullary and cortical cells lack pigment granules entirely, some have a full complement, and some have a reduced number of granules. These various cells are arranged irregularly throughout the length of the hair. With increasing size of hairs (such as awls in the mouse, most of the hairs in rabbits and cats, all of the hairs in guinea pigs), the follicles are less "sensitive" to a given dose of x-radiation, produce a greater frequency of mosaics, and display less difference in response when treated in telogen or in anagen phases. It would seem that susceptible elements must be few in number, very few in smaller follicles. At beginning of anagen there would be a moderate increase of these elements but little, if any, further increase during the anagen phase (2 to 17 days post-plucking). After the initial supply for the original follicle invagination, there would be no new invasion for subsequent hair generations.

Following Masson's (1948) definitions the dendritic cells which eventually produce pigment are termed melanoblasts whereas the recipient cells, if any, are termed melanophores. Melanoblasts generally are not observed because of their fragility with certain standard histological methods and because they are largely obscured after the matrix cells become pigmented. Phase microscopy with the Spencer B minus contrast low, oil immersion objective on unstained frozen sections has proved most revealing. In young mice, melanoblasts of the basal layer of the skin epidermis can be seen to be incorporated in the original invaginations of newly-forming follicles. In early anagen stages of subsequent hair generations, melanoblasts are found in the permanent external sheath (continuous with basal layer of skin epidermis) or in the derivative basal layer of the bulb. They become melanogenic with fine dispersed granules and send long dendritic processes to the matrix cells. Later, beginning about 6 days post-plucking, the "inoc-